Chapter 4

Therapy evaluation of laryngeal carcinomas by Tyrosine-PET

Jurjan R. de Boer [1], Jan Pruim [5], Fred Burlage [4], Allard Krikke [3], Anton T.M.G. Tiebosch [2], Frans W.J. Albers [1], Willem Vaalburg [5], Bernard F.A.M. van der Laan [1]

Departments of [1] Otorhinolaryngology and Head and Neck Surgery, [2] Pathology, [3] Radiology, [4] Radiotherapy and [5] PET Center of University Hospital Groningen, the Netherlands

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INTRODUCTION

Treatment of laryngeal squamous cell carcinomas consists primarily of radiotherapy, either as single therapy or in combination with surgery. Radiotherapy is preferred over surgery based on the preservation of laryngeal function, including the voice, and therefore preserving quality of life. However, radiotherapy may cause a variety of acute and chronic tissue effects and anatomic distortion such as fibrosis, edema, inflammation and scarring [1]. The chronic effects can last up to 1½ years after primary treatment before diminishing in severity. Post-treatment surveillance consists of frequent physical examinations, indirect laryngoscopy, and in case of clinical suspicion, radiological imaging (CT/MRI) and endoscopic examination under general anesthesia with biopsies of suspicious areas.

Local recurrent disease occurs in up to 50% of patients with T2-T3 laryngeal squamous cell carcinomas [2], and in approximately 90% of the cases, this recurrence will develop within the first 2 years after radiotherapy. Differentiation between therapy induced tissue changes and residual or recurrent disease by clinical examination or conventional imaging is difficult in the coinciding period after treatment. The findings on CT and MRI are often equivocal in the differentiation of benign posttreatment reactive tissue reactions versus viable tumor [3].

Only histology obtained by biopsy proves residual or recurrent disease but may cause false-negative results by missing submucosal tumor growth [4]. Deep or repeated biopsies under general anaesthesia are often needed but should be performed with caution, because the capacity of irradiated tissue to recover is diminished [5], and voice quality may be compromised. Because of these diagnostic limitations, delay in detection of local recurrent disease may occur, with consequent effects on survival and morbidity [6]. Therefore, non-invasive methods for accurate assessment of therapy response and early detection of recurrence will probably be an important tool for everyday clinical practice.

Positron emission tomography (PET) is a functional imaging modality that enables determination of tissue metabolism and pathophysiology in vivo and, therefore, of tumor tissue metabolism. In contrast to CT and MRI, PET is not hampered by anatomical or structural changes, because it reflects metabolism and alterations therein. The glucose analogue 2-[^{18}F]-fluoro-2-deoxy-D-glucose (FDG) is the most widely used radiopharmaceutical in PET. FDG is an indicator of tumor metabolism based on increased glycolysis in tumor cells [7]. The application of FDG-PET is successful for a variety of malignancies [8], including primary head and neck cancers and metastatic cervical lymph nodes [9,10]. Unfortunately, FDG also accumulates in inflammatory tissue, which may cause false-positive results [11]. In the first 1½ year after radiotherapy, this significant drawback may reduce the specificity of FDG-PET [12]. Consequently, a search for alternative and more specific tracers is ongoing.

Amino acids are less avidly metabolized by inflammatory cells and therefore ^{11}C-labeled amino acids were introduced as radiotracers [13,14]. It was also demonstrated that amino acid uptake in tumor tissue is high compared with normal tissue because of an

increased protein synthesis [15]. Of the amino acids available (both native and artificial), methyl-labeled ^{11}C-methionine (MET) is the most frequently used, mainly because of the relative ease of synthesis. However, the position of the ^{11}C-label does not allow for quantitative studies [14]. In contrast, L-[1-^{11}C]-tyrosine (TYR), a carboxyl-labeled amino acid, is an appropriate compound to determine protein synthesis activity in tumor tissue.[16] By using a dynamic scanning procedure, calculation of the Protein Synthesis Rate (PSR) of tumor tissue is possible. TYR-PET has been successfully used in detection and quantification of a variety of primary and recurrent tumors [17-20].

The primary aim of this study was to investigate the potential of dynamic TYR-PET for therapy evaluation in patients with laryngeal cancer. The diagnostic accuracy for detection of residual or recurrent disease was analyzed and compared to conventional methods. Also, different methods for quantification of metabolic activity after primary radiotherapy of laryngeal carcinomas were assessed and compared.

PATIENTS AND METHODS

Patients

Nineteen patients, 2 women and 17 men (median age 59; range 45-81 years), with T2-T3 laryngeal squamous cell carcinomas suitable for definitive radiotherapy were included (Table 1). All patients underwent physical examination of the head and neck, TNM-staging (UICC,1997) [21] including laryngoscopy under general anaesthesia and biopsies of suspicious areas, and CT imaging before final therapy. Primary laryngeal tumors were localized in the glottic area in 58% (n = 11) and in the supraglottic area in 42% (n = 8). All but one patient had T2 disease, and three patients had lymph node metastases (N1 in two, N2 in one). In addition to the current diagnostic modalities, dynamic TYR-PET imaging (PET1) was performed before definitive therapy, according to the procedure outlined in the following.

All patients received megavoltage radiation with a conventional fractionation schedule to a total absorbed tumor dose of 70 Gy, 2 Gy per fraction, five fractions weekly.

Three months after radiotherapy, all patients had a second TYR-PET scan (PET2). If residual disease was suspected clinically, additional CT imaging and biopsy verification of disease status were performed. Residual tumor was diagnosed if persistent disease was confirmed within six months after radiotherapy.

The minimal follow-up period after the first TYR-PET was 29 months (range 29-52 months). If recurrent tumor was clinically suspected during follow-up, a third TYR-PET scan (PET3) and additional CT and biopsy were indicated. Persistent or increased edema; impaired vocal cord mobility; local ulceration; and/or persistent complaints of pain, otalgia, and swallowing problems were reasons for suspecting local recurrence. Recurrent

disease was diagnosed if progression of disease was confirmed histologically later than six months after radiotherapy. If residual or recurrent disease was confirmed by biopsy, patients were scheduled for total laryngectomy.

The study protocol was approved by the Medical Ethics Committee of the Groningen University Hospital, and written informed consent was given by each patient.

Table 1
Patient and tumor characteristics

Characteristics		Number
Men		17
Women		2
Primary site larynx		
	-glottic	11
	-supraglottic	8
Primary tumor		
T2		18
T3		1
Lymph nodes		
N0		16
N1		2
N2		1
N3		0
UICC stage		
I		0
II		15
III		3
IV		1

UICC = International Union Against Cancer (1997)

CT imaging

CT scans were performed on a Volume Zoom CT scanner (Siemens, Erlangen, Germany). Intravenous contrast was given in all studies as bolus of 100-120 ml. Transverse 5-mm consecutive sections were obtained from the mastoid tip to the clavicles. CT scans were considered positive for malignancy in case of an abnormal mass with enhancement or in case of soft tissue increase compared with previous CT scans. Sensitivity and specificity of CT for detection of residual or recurrent disease were calculated.

TYR-PET

In all 19 patients, dynamic TYR-PET studies were performed at the time points outlined previously. TYR was produced by means of a modified microwave-induced Bücherer-Strecker synthesis [22] with a radiochemical purity greater than 99%. Studies were acquired using an ECAT 951/31 PET camera (Siemens/CTI, Knoxville, TN). This device has a 56-cm-

diameter patient aperture and acquires 31 planes simultaneously over a 10.8-cm axial field of view.

Patiënt refrained from food intake for at least eight hours before the investigation, but were allowed to drink noncaloric beverages and to use their normal medication. A venous canula was placed in the anticubital vein of the forearm for injection of TYR. The injected dose varied from 144 to 377 MBq (median 366 MBq). An arterial canula was inserted under local anesthesia into the radial artery of the contralateral arm for sampling arterial blood during data acquisition. The head of the patient was fixed with the Frankfurter horizontal plane (line between the external meatus acusticus and the lower orbital rim), making an angle of 110° with the horizontal bed position. Before sampling, the nonradioactive tyrosine concentration in plasma was assessed. In the arterial blood samples, plasma activity of TYR, $^{11}CO_2$ and ^{11}C-protein levels were measured by radio-HPLC [16].

A transmission scan to correct for photon attenuation by body tissues in the imaged area was obtained immediately before the emission scan. Dynamic scanning with 16 time frames was performed from the time of injection to 50 min after injection at the level of the tumor. The protocol included ten frames of 30 sec, three frames of 5 min and three frames of 10 min. Image data were backprojected using a 0.5 cycle/pixel Hann-filter, which yielded a resolution of 6 mm full-width-of-half-maximum (FWHM).

PET data

Visual analysis

PET images were displayed in coronal, sagittal and transaxial projections on a computer display using standard ECAT software and interpreted independently by two experienced physicians. Visual analysis was graded as positive when high TYR uptake was observed as a focal hot spot on a nonphysiological location. Otherwise, the image was interpreted as negative for malignancy. Sensitivity and specificity of TYR-PET for detection of residual or recurrent disease were calculated.

Quantitative analysis

Absolute quantification was performed by assessment of tumor PSR. The PSR was obtained by placing a region of interest (ROI) in the plane with most intense uptake at the site of the tumor as observed at visual analysis, using a 70% threshold of maximum intensity. The tissue time-activity curve obtained from this ROI, together with the plasma-input data (MBq/ml TYR corrected for $^{11}CO_2$ and ^{11}C-labeled plasma proteins) were used to calculate PSR in nanomoles per milliliter tumor tissue per minute (nmol/ml/min) using a modified Patlak analysis [16]. By masking nontumor regions with physiologically high uptake of TYR (e.g., parotid glands), spillover of these regions to the average time-activity curve was prevented.

To compare absolute quantification and semiquantitative methods, Standardized Uptake Values (SUV) were calculated from the summed data obtained from the last three frames (20-50 min after injection). SUVs were calculated by dividing tissue activity (MBq/ml) by injected dose (MBq) based on BW (SUV_{BW}) or LBM (SUV_{LBM}).[23] In addition, the tumor-to-nontumor ratio (T/N) was calculated.

Statistical analysis

The quantitative values of tumor tissue compared to the corresponding normal tissue were analyzed by the Wilcoxon test. Mann-Whitney U test was used to compare differences in PSR, SUV, $PSR_{T/N}$ and T/N. Correlations between quantitative values were evaluated by the two-tailed Pearson test. In these tests, a p-value of < 0.05 was considered to be statistically significant.

RESULTS

Pretreatment results

Nineteen TYR-PET studies were performed before definitive radiotherapy (Table 2). All histologically proven laryngeal carcinomas were depicted by TYR-PET and CT. The PSR of tumors ranged from 0.87 to 3.30 nmol/ml/min (median 1.95 nmol/ml/min), which differed significantly ($p<0.001$, Wilcoxon) from the PSR of normal tissue (median 0.51; range 0.22-0.77). The median SUV_{BW} (range 1.85-5.88), SUV_{LBM} (range 1.39-3.99) and T/N-ratio (range 2.20-5.81) were 4.20, 2.89 and 4.33, respectively. Significant correlations ($p<0.001$) were observed between PSR and SUV_{BW} ($r = 0.76$) and SUV_{LBM} ($r = 0.82$).

Evaluation of response to radiotherapy

A second TYR-PET scan (PET2), performed three months after irradiation for evaluation of therapy, was possible in 15 of the 19 patients (Table 2). Four patients did not complete follow-up because of patient-related reasons. Of the remaining fiveteen patients, 7 were suspected of having residual disease by physical examination at the time of scanning. In addition to the second TYR-PET scan, CT imaging and biopsies during laryngoscopy under general anaesthesia were performed in these 7 cases. Of the patients without clinical suspicion of residual disease by physical examination ($n = 8$), only a second TYR-PET scan was performed.

Visual analysis

Of the 7 patients clinically suspected of having residual disease, TYR-PET was positive in 4 cases. Residual disease was histologically confirmed in these four patients, and therefore

Table 2

Results of Physical examination, Computed Tomography, Histology and Positron Emission Tomography before radiotherapy, 3 months after radiotherapy (RTX) and during follow-up.

Patient	Pretreatment					Posttreatment					Follow-up					Outcome
nr/sex/age	PE	CT	PA	PET	PSR	PE	CT	PA	PET	PSR	PE	CT	PA	PET	PSR	
1/M/79	+	+	+	+	2.50				NA							
2/M/44	+	+	+	+	2.40				NA							
3/M/68	+	+	+	+	1.62	+	+	-	-	0.72						NED
4/M/64	+	+	+	+	2.06	-			-	0.64	+	-	+	+	0.93	Recurrent
5/M/68	+	+	+	+	2.19	-			-	0.60	+	-	-	-	0.64	NED
6/F/60	+	+	+	+	2.68	+	+	-	+	0.93	+	+	+	+	0.83	Residual
7/M/64	+	+	+	+	1.44	-			-	0.74						NED
8/M/66	+	+	+	+	0.87	-			-	0.49						NED
9/M/54	+	+	+	+	1.58	-			+	0.55	+	+	+	+	1.40	Recurrent
10/M/46	+	+	+	+	1.51	-			-	0.68						NED
11/M/45	+	+	+	+	1.66	+	-	+	+	1.29						Residual
12/M/56	+	+	+	+	2.48				NA							
13/M/59	+	+	+	+	1.95	-			-	0.57						NED
14/M/65	+	+	+	+	3.30	+	-	-	+	1.34	+	+	+	+	1.85	Recurrent
15/M/77	+	+	+	+	1.14				NA							
16/M/52	+	+	+	+	3.14	+	-	-	-	0.76	+	+	-	-	0.82	NED
17/F/59	+	+	+	+	1.35	-			-	0.88						NED
18/M/54	+	+	+	+	1.43	+	-	-	-	0.93						NED
19/M/46	+	+	+	+	2.28	+	+	+	+	2.42						Residual

Abbreviations: + or - : presence or absence of cancer by investigation modality; PE = Physical Examination; CT = Computed Tomography; PA = Pathology; PET = Positron Emission Tomography; PSR = Protein Synthesis Rate (in nmol/ml/min); NA = Not Available; NED = No Evidence of Disease

the scans were scored as true-positives. The three negative TYR-PET scans were scored true-negative, because repeated biopsies by laryngoscopy under general anesthesia maintained negative, and progression of disease was not found during follow-up.

Of the 8 patients without clinical suspicion of residual disease, seven TYR-PET scans were negative and one was positive. TYR-PET proved to be true-positive in this case because within six months after radiotherapy, progression of disease was histologically confirmed. The seven TYR-PET examinations assessed negative for residual tumor, and for which confirmatory biopsies were not performed, were all scored true-negatives, because progression of disease did not occur within six months after radiotherapy.

A sensitivity and specificity of 100% for TYR-PET in determination of tumor status after radiotherapy was calculated. Positive and negative predictive values were also 100%. Sensitivity and specificity were 80% and 70% for physical examination and 50% and 67% for CT, respectively.

Quantitative analysis

Quantitative PET analysis was performed on all 15 posttreatment TYR-PET scans (Table 3). The median PSR of all subjects was 0.74 nmol/ml/min (range 0.49-2.42 nmol/ml/min) after therapy, which was a significant decrease ($p<0.001$) when compared to pretreatment PSR. Significant decreases ($p<0.001$) were also observed in SUV_{BW} , SUV_{LBM} and T/N-ratio. No correlation was found after radiotherapy between PSR and SUV_{BW}, and only weak correlation ($r = 0.56$) between PSR and SUV_{LBM}.

Table 3

Quantitative PET analysis preradiotherapy and postradiotherapy.

Median and range of PSR, SUVs and T/N-ratio of TYR-PET before radiotherapy (PET1) and 3 months after radiotherapy (PET2). Values of true-positive (TP) and true-negative (TN) scans 3 months after radiotherapy are listed.

	Pretreatment PET (PET1)	Posttreatment PET (PET2)
	Median (Range)	Median (Range)
PSR	1.95 (0.87-3.30)	0.74 (0.49-2.42) [(a)]
TP		1.29 (0.55-2.42) [(1)]
TN		0.70 (0.49-0.93)
SUV_{BW}	4.20 (1.85-5.88)	2.36 (1.20-3.55) [(b)]
TP		2.67 (2.36-3.55) [(2)]
TN		2.17 (1.20-2.53)
SUV_{LBM}	2.89 (1.39-3.99)	1.56 (0.94-2.82) [(c)]
TP		2.05 (1.71-2.82) [(3)]
TN		1.52 (0.94-1.84)
T/N-ratio	4.33 (2.20-5.81)	1.61 (0.69-3.82) [(d)]
TP		2.89 (2.01-3.82) [(4)]
TN		1.43 (0.69-1.68)

Significant differences in pre- and posttreatment [(a)] PSR ($p<0.001$), [(b)] SUV_{BW} ($p<0.001$), [(c)] SUV_{LBM} ($p<0.001$) and [(d)] T/N-ratio ($p<0.001$).
Differences between TP and TN are [(1)] $p=0.057$, [(2)] $p=0.013$, [(3)] $p=0.003$ and [(4)] $p=0.001$

If quantitative values of residual disease and benign posttherapy tissue were compared, no significant differences in PSRs ($p=0.057$) were found between true-positive (median 1.29; range 0.55-2.42) and true-negative scans (median 0.70; range 0.49-0.93). However, SUV_{BW} ($p=0.013$), SUV_{LBM} ($p=0.003$) and T/N-ratio ($p=0.001$) differed significantly

between true-positive and true-negative scans (Table 3). Only in the T/N-ratio values were no overlap between true-positive and true-negative scans observed (Fig. 1).

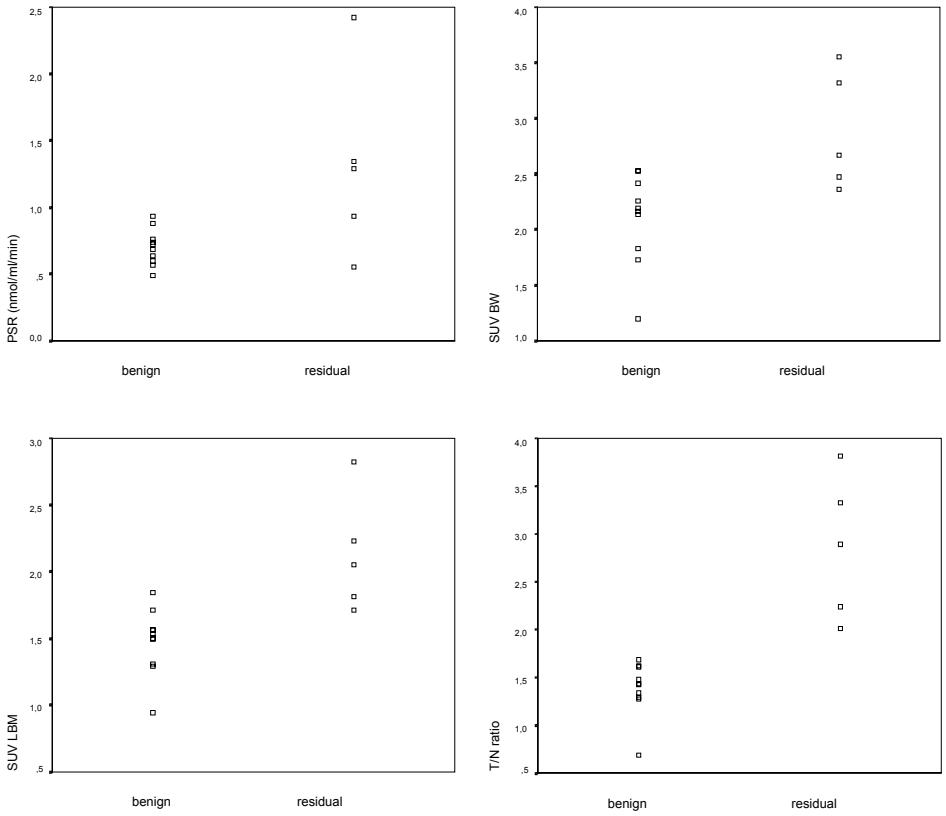

Figure 1.
L-[1-^{11}C]-Tyrosine uptake 3 months after radiotherapy (PET2) in residual and benign posttreatment tissue presented as PSR, SUV_{BW}, SUV_{LBM} and T/N-ratio.

Detection of recurrent tumor

After the second TYR-PET, patients had clinical follow-up for at least 26 months. During this follow-up period, six patients were suspected of having recurrent disease. CT imaging, biopsies and a third TYR-PET scan (PET3) were performed in all six cases (Table 2).

Visual analysis

TYR-PET scans were positive for recurrent disease in four patients, which were all histologically confirmed. The two other PET scans were negative, and in these cases no evidence of disease was found during follow-up (at least 12 months after PET3). Therefore, sensitivity and specificity of TYR-PET for detection of recurrent primary tumor are 100% (positive and negative predictive values, 100%). Sensitivity and specificity of CT in detecting

recurrent malignancy were 75% and 50%, respectively, with one false positive and one false negative result. Physical examination proved 100% sensitive, but 0% specific. Figure 2 shows an example of consecutive TYR-PET scans of a patient with good response to radiotherapy after 3 months but recurrent disease within 18 months after treatment.

Figure 2.
Consecutive TYR-PET scans of a T2 N0 laryngeal carcinoma (patient 4) with increased uptake pretreatment (PET1), good response to radiotherapy after 3 months (PET2), but recurrent disease 18 months after therapy (PET3).

Quantitative analysis
In all six patients with suspicion of recurrent disease, quantitative analysis was performed. Median and range of PSR, SUV and T/N-ratio are mentioned in Table 4. Although statistical analysis was not valid because of the limited number of patients with recurrent disease, in true-positive PET scans a higher PSR (median 1.17; range 0.83-1.85) was measured as compared with true-negative scans (median 0.73; range 0.64-0.82). These differences were also found between true-positive and true-negative values of SUV_{BW}, SUV_{LBM} and T/N-ratio (Table 4). No overlap was observed between true-positive and true-negative values in all quantitative methods (Fig. 3).

DISCUSSION

Monitoring the response to radiotherapy is one of the major challenges in head and neck oncology. After irradiation, evaluation for residual or recurrent disease by physical examination and conventional imaging may be hampered by posttreatment fibrosis, edema, inflammation or scarring. These factors limit the sensitivity and specificity of interpretation of anatomical imaging modalities, such as CT and MRI [3].

PET, which assesses abnormal metabolic activity of tumors rather than anatomical changes, avoids some of the difficulties inherent in examining the posttreatment head and neck. The feasibility of FDG-PET in the identification of viable tumor in head and neck after treatment has been extensively studied. FDG-PET showed comparable or better sensitivity and specificity than CT or MRI for detection of locoregional recurrence. A study by Bailet et al.[9] in which PET and MRI were used to evaluate 10 patients within 4 months after radiotherapy, showed a distinct superiority of PET over MRI. Anzai et al.[24] found

sensitivities and specificities of 88% and 100% for FDG-PET, and 25% and 75% for CT/MRI, respectively, in 12 patients with a history of recurrent head and neck carcinoma.
In most FDG studies that included 10 to 15 patients, sensitivity for detection of residual

Table 4
Quantitative analysis of recurrent disease.
Median (range) of PSR, SUVs and T/N-ratio of TYR-PET scans three months after radiotherapy (PET2) and during follow-up (PET3) are listed, subdivided in true-positive (TP) and true-negative (TN) values for PET3.

	Posttreatment PET (PET2) (3 months after RTX)	Follow-up PET (PET3)
	Median (Range)	Median (Range)
PSR	0.74 (0.49-2.42)	0.88 (0.64-1.85)
TP		1.16 (0.83-1.85)
TN		0.73 (0.64-0.82)
SUV_{BW}	2.36 (1.20-3.55)	3.03 (1.34-4.77)
TP		3.36 (2.70-4.77)
TN		1.84 (1.34-2.33)
SUV_{LBM}	1.56 (0.94-2.82)	2.26 (0.97-2.70)
TP		2.46 (2.08-2.70)
TN		1.34 (0.97-1.71)
T/N-ratio	1.61 (0.69-3.82)	2.71 (0.82-3.92)
TP		3.53 (2.16-3.92)
TN		1.15 (0.82-1.47)

or recurrent tumor ranged from 88% to 100%, while specificity could be as low as 43% [10,25,26]. More recent FDG-PET series with larger number of patients reported sensitivities ranging from 50% to 100% and specificities from 83% to 95% [27,28]. Only one study reported sensitivity of 100% and specificity of 100% for FDG-PET compared with 75% and 80 % for CT/MRI in detection of recurrent tumor [29].

The variation in specificity of FDG-PET is caused by the number of false-positive results after irradiation because of the increased glucose metabolism in tissues other than malignancies. Several authors reported false-positive findings in their series in which histopathologic examination showed chronic postradiation inflammation changes without evidence of recurrence [24,28,30]. In a preclinical study the FDG uptake in mouse tumors was investigated with microautoradiography, and it was found that twenty-nine

percent of the glucose utilization took place in nontumor macrophages that were located in the necrotic areas of the tumor [31]. Strauss et al. reviewed the differentiation of malignant and benign lesions by FDG-PET and confirmed the relatively low specificity of FDG [11].

Amino acids were introduced as radiopharmaceuticals in order to provide an alternative to this, and demonstrated high uptake in tumor tissue due to an increased protein synthesis [15]. In contrast to glucose, amino acids are less avidly metabolized in inflammatory tissue, which may be advantageous when applying PET for therapy evaluation [13,14].

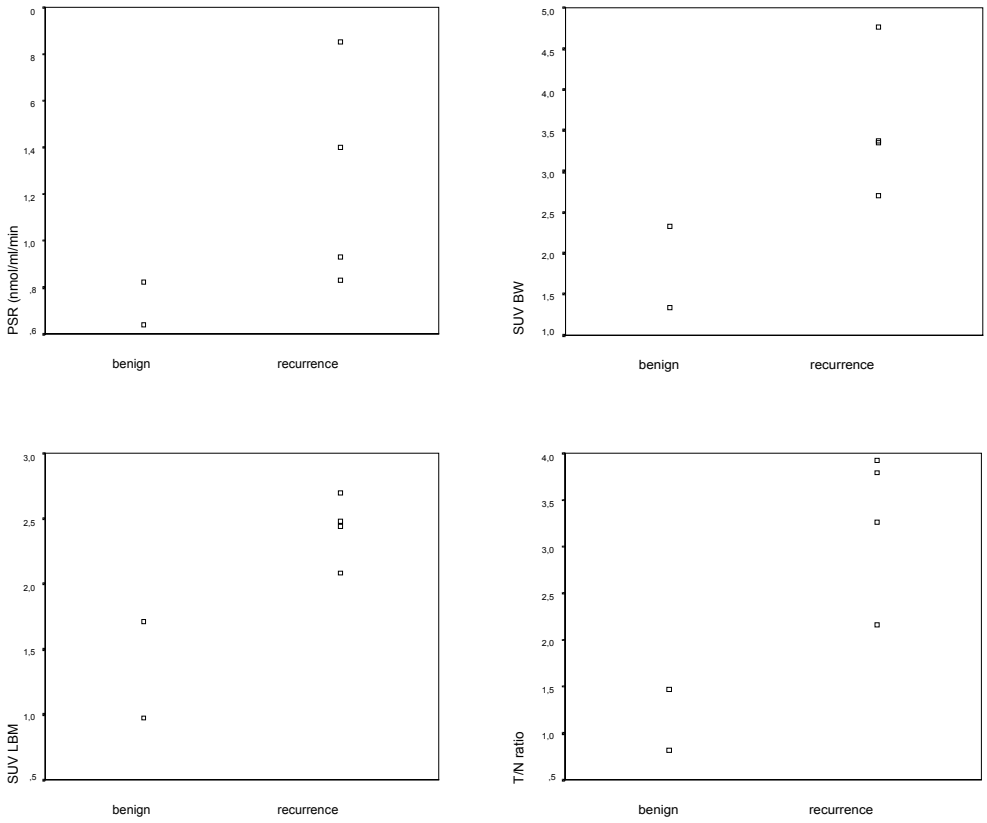

Figure 3.

Tracer uptake in recurrent and benign posttreatment tissue during follow-up (PET3) presented as PSR, SUV_{BW}, SUV_{LBM} and T/N-ratio.

The most frequently used radiolabeled amino acid is L-[methyl-^{11}C]-methionine (MET). The Turku University group has extensively investigated head and neck cancer with MET-PET. A therapy evaluation study by Lindholm et al. including 15 patients, demonstrated significant differences in SUVs between complete responders to radiotherapy and patients with persistent disease [32]. However, in the study by Nuutinen et al., no differences were observed after treatment between quantitative values of responding and relapsing patients

[33]. Furthermore, it was demonstrated that quantification of protein synthesis with MET is rather difficult. MET is involved in several metabolic pathways, such as transmethylation and polyamine synthesis, and is converted in S-adenosyl methionine. This may lead to the accumulation of nonprotein metabolites in tumor tissue. The complicated metabolism of methyl-labeled methionine has made it impossible to construct a precise metabolic model with consequent effect on quantification [14,34].

Carboxyl-labeled amino acids, such as L-[1-^{11}C]-tyrosine (TYR), seem to be more appropriate compounds to assess protein synthesis activity in tumor tissue [35]. TYR-PET has been successfully used in the detection and quantification of PSR in various primary and recurrent tumors and proved superior to FDG in some studies [16-18,20,36]. Van Ginkel et al. used TYR-PET successfully to evaluate the effect of chemotherapy in patients with soft tissue sarcomas and skin cancer. High sensitivity (82%) and specificity (100%) of TYR-PET were found in detection of relapsing disease. Significant differences in PSR values were observed between patients with complete, partial or no response to therapy [19].

In this study, TYR-PET was highly sensitive and highly specific for tumor status after radiotherapy and also for detection of residual or recurrent disease. These figures proved favorable in comparison to CT imaging and physical examination. We observed no false positive or false negative results by TYR-PET performed three months after completion of radiotherapy. In FDG-PET studies, false negative results were described in scans performed earlier than four months after radiation therapy [25]. The predictive values of TYR-PET three months after radiotherapy and during follow-up were all 100%. The high positive predictive value is useful in early detection of residual or recurrent disease. In the evaluation of response three months after therapy, the sensitivity of biopsies for confirmation of residual disease was 50%, if a follow-up of six months for presence or absence of tumor was considered. Repeated biopsies were necessary in two patients before residual tumor was proven. TYR-PET proved to be more sensitive for detection of residual disease in comparison with histological confirmation by biopsy.

Although the high positive and negative predictive values make TYR-PET a strong diagnostic tool in monitoring response to therapy and early detection of recurrent tumor, the number of cases and the variance in tumor stage are limited. Assessment of the accuracy of TYR in a larger group of patients with laryngeal carcinomas is necessary, because non-tumoral uptake of amino acids has been described [14]. Future studies have to determine whether the sensitivity, specificity and negative predictive value of TYR-PET may prevent unnecessary diagnostic procedures that result from indeterminate clinical or imaging findings in patients treated for squamous cell carcinomas of the head and neck.

Absolute quantification was performed in this study by determination of PSR. Three months after treatment, significant decrease in PSR was found of all tumors compared to pretreatment PSR. However, near significant ($p=0.057$) differences in PSR values were observed between residual disease and benign posttherapy tissue. Overlap of PSR values was found in two cases (Fig. 1).

Although determination of PSR is the most preferable form of quantification, it

requires elaborate protocols with dynamic scanning and arterial cannulation to obtain blood samples for the tissue input function. Such protocols are cumbersome for the patient and therefore often replaced by the relatively simple calculation of SUV. This calculation only requires a short attenuation-corrected static scan, without blood sampling and therefore is more patient friendly. In this study, quantification of tumor activity by SUV_{BW}, SUV_{LBM} and T/N-ratio was performed. Significant differences were found between residual disease and benign posttherapy tissue in SUV_{BW} (p=0.013), SUV_{LBM} (p=0.003) and T/N-ratio (p=0.001). Only the T/N-ratio showed no overlap in values and differentiation could be determined between benign tissue (T/N-ratio < 2.0) and residual disease (T/N-ratio > 2.0). After treatment, only weak correlations were observed between PSR and the semiquantitative methods. Although we do not have a plausible explanation for this, the strong discrimination capacity of SUVs or T/N-ratio make them useful methods for quantification during evaluation of therapy.

Quantification of recurrent disease during follow-up was also performed by calculation of PSR, SUV_{BW}, SUV_{LBM} and T/N-ratio. Although no statistical analysis was possible, all four methods demonstrated distinct differences between benign lesions and recurrent disease, and no overlap in values was found. Further investigations have to be performed to reveal the most accurate quantification method for tumor recurrence.

Because we found no differences in visual analysis and quantitative analysis by PSR, SUV_{BW}, SUV_{LBM} or T/N ratio, visual interpretation of TYR-PET seems a sufficiently adequate method to assess residual or recurrent disease. However, some studies demonstrated that due to radiotherapy, tumor and background tissues are likely to change in metabolic rate and visual discrimination may become difficult [37,38]. In such cases quantitative methods may prove more accurate than visual interpretation, although our study does not confirm this statement. In equivocal PET scans, differentiation between benign posttherapy tissue and viable tumor on quantitative values may prove advantageous. Furthermore, quantification of tumors may have prognostic value which is currently under investigation.

CONCLUSIONS

TYR-PET is a powerful diagnostic imaging modality in the evaluation of response to therapy and detection of recurrent disease in laryngeal squamous cell carcinomas. Although the number of patients was limited, PET proved more sensitive (100%) and specific (100%) than conventional imaging and FDG-PET for detection of residual or recurrent disease. The high positive predictive value of TYR-PET is useful in early detection of residual or recurrent disease.

In evaluation of therapy, absolute quantification by PSR gave similar results as quantification by SUV_{BW}, SUV_{LBM} and T/N ratio. However, no differences in quantitative analysis and visual analysis were observed in this study, and therefore visual interpretation of TYR-PET scans seems sufficiently adequate for therapy evaluation.

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